Primer

The complement system

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The system of plasma proteins called complement was so-named because it complements the activity of antibody in the lysis of bacteria. It has a central role in host defence against many micro-organisms and in the modulation of inflammatory reactions, an activity that has been illuminated by the study of humans with naturally occurring deficiencies of complement.

Complement is activated by three pathways: the classical, alternative and mannan-binding lectin (MBL) pathways (Figure 1 and Table 1). The classical pathway is primarily activated by antibody bound to antigens (immune complexes). The alternative pathway is activated by the direct binding of complement component C3 to pathogens in the absence of antibody. The recently discovered MBL pathway is initiated by the binding of MBL to mannose groups, present on many bacterial cell walls. All three pathways lead to the cleavage of C3, the central component of complement, which functions as an opsonin, tagging pathogens and immune complexes for recognition and uptake, mediated by specific complement receptors on phagocytic cells. Subsequent activation of the terminal complement pathway, known as the membrane attack complex (MAC), leads to cell lysis.

An important current focus of complement research is the physiological activities of complement *in vivo*, on which we will concentrate in this primer.

Complement and inflammation

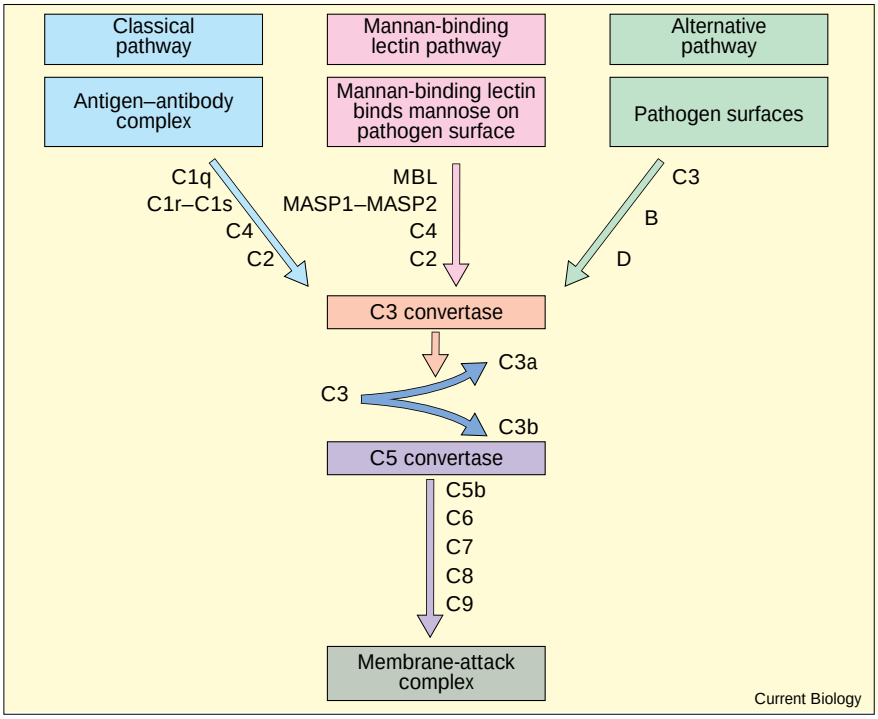
Inflammation is a complex process involving vasodilation, increased vascular permeability and extravasation of plasma. This is accompanied by the adhesion of leukocytes to vascular endothelium and their emigration into surrounding tissue. Complement plays an important part in inflammation, in helping to recruit effector cells and in promoting the killing and clearance of pathogens.

But complement can be both friend and foe. In some circumstances the activation of complement in response to immune complexes may cause harm: acutely in the context of massive complement activation occurring in patients with overwhelming Gram-negative bacterial sepsis, and chronically, when an antibody response develops in the context of antigens that cannot be cleared. Examples of this are infections at anatomically-protected sites — such as heart valves in bacterial endocarditis — or infections by certain viruses, such as hepatitis C, in patients who are unable to clear the infection. The second category in which there is persistent formation of immune complexes is autoimmune disease, in which, by definition, the antigen is a permanent feature of the host.

An understanding of how immune complexes cause tissue injury developed from study of such diseases in humans and animals, and from the induction of experimental immune complex disease. In these situations, immune complexes were shown to cause tissue injury through activation of complement, as opsonin (binding to complement receptors on leukocytes) and as the source of anaphylatoxins (stimulating leukocyte chemotaxis and degranulation). But the development of gene-targeted mice with deficiencies of complement proteins and of receptors for the Fc part of antibodies has led to re-appraisal of the role of complement and antibodies in inflammatory injury.

Some studies of the formation of immune complexes in the skin of these animals (the Arthus reaction)

Figure 1



The main pathways and components of the complement activation system.

Table 1

Effector activities of the complement system

Activity	Complement protein	Complement receptor mediating effect
Opsonin for phagocytosis	C3b, iC3b C4b C1q, MBL	CR1, CR3 CR1 C1q receptor
Peptide mediators of inflammation	C5a C3a	C5a receptor C3a receptor
Lytic membrane attack complex	C5b, C6, C7, C8, C9	
Processing and clearance of antigen–antibody immune complexes	C1q C3b, iC3b C4b	C1q receptor CR1, CR3 CR1
Localization of antigen in the form of immune complexes on antigen-presenting cells	C3b, iC3b, C3dg	CR1, CR2, CR3
Activation of B lymphocytes	C3dg	CR2
Clearance of apoptotic cells	C1q?	C1q receptor?

The activites of the complement system are shown in the left-hand column. In the centre are shown the complement proteins that mediate the effects. (C3b, iC3b and C4b are large activation products of these complement proteins that bind covalently to the targets of complement activation.) The column on the right shows the receptors that are bound by the relevant complement ligands in the centre column.

showed that ligation of Fc receptors — which bind to antibodies complexed with antigens — on mast cells is critical for the induction of inflammatory injury by immune complexes, and that complement plays little or no part. This view has been supported by the study of the effects of Fc receptor deficiency in a spontaneous mouse model of an autoimmune disease, systemic lupus erythematosus, in which immune complexes are prominent. Fc receptors were shown to be required for the development of severe renal disease.

It is probably too early, however, to dismiss completely the role of complement in inflammation in immune complex-mediated disease. Studies of immune complex formation in mice deficient in complement component C5, or in the receptor for the anaphylatoxin C5a, demonstrated a more substantial role for complement in the mediation of inflammatory reactions in the lungs and peritoneum. Similar results were obtained in studies of peritoneal infection in C3- and C4-deficient mice. Complement activation was shown to mediate the initial release of the pro-inflammatory cytokine TNFα by mast cells, and the subsequent neutrophil influx.

Complement and adaptive immunity

It has been known since the seminal experiments of Pepys in 1974 that complement is involved in the induction and maintenance of antibody responses. Recently, there has been an increasing appreciation of the importance of the role of complement at the interface between innate and adaptive immunity. Here, complement plays a part in the localization of antigens to antigen-presenting cells — cells specialized to present antigens to lymphocytes and to activate them — and to B cells, both of which bear receptors for major cleavage products of C3.

This activity of complement was beautifully demonstrated by experiments in which antibody responses were measured in animals immunized with antigens covalently linked by genetic engineering to the relevant cleavage product of C3. The addition of increasing numbers of C3 molecules to each antigen molecule dramatically reduced the threshold for activation of B lymphocytes.

Evasion of complement by pathogens

One of the most fascinating aspects of the study of complement is the exploration of the mechanisms that pathogens have evolved to evade and, in some cases, exploit the complement system. Pathogens that cause disease are pathogenic by virtue of their ability to escape, at least in part, the primary mechanisms of defence of the innate immune system. To evade complement pathogens have stolen and mimicked regulatory molecules of the complement system (Figure 2).

Complement and autoimmunity

Homozygous deficiency of classical pathway proteins is very strongly associated with the development of the autoimmune disease systemic lupus erythematosus (SLE). There is a hierarchy of susceptibility and severity of autoimmune disease according to the position of the deficient protein in the classical pathway, with C1q deficiency showing the highest prevalence and causing the most severe disease. These clinical observations imply that there is a, so far unknown, physiological activity of the early proteins of the classical pathway that protects against the development of SLE.

Some recent data point to the discovery of a possible new physiological activity of the complement system that could link complement deficiency and SLE. There is evidence that apoptotic cells might be the source of the autoantigens that stimulate autoimmunity in SLE and that C1q might be involved in the processing of apoptotic cells. If these findings are confirmed, then failure of clearance of apoptotic cells could perhaps explain the link between complement deficiency and SLE. The development of C1q-deficient mice will help to resolve this.

Therapeutic inhibition of complement

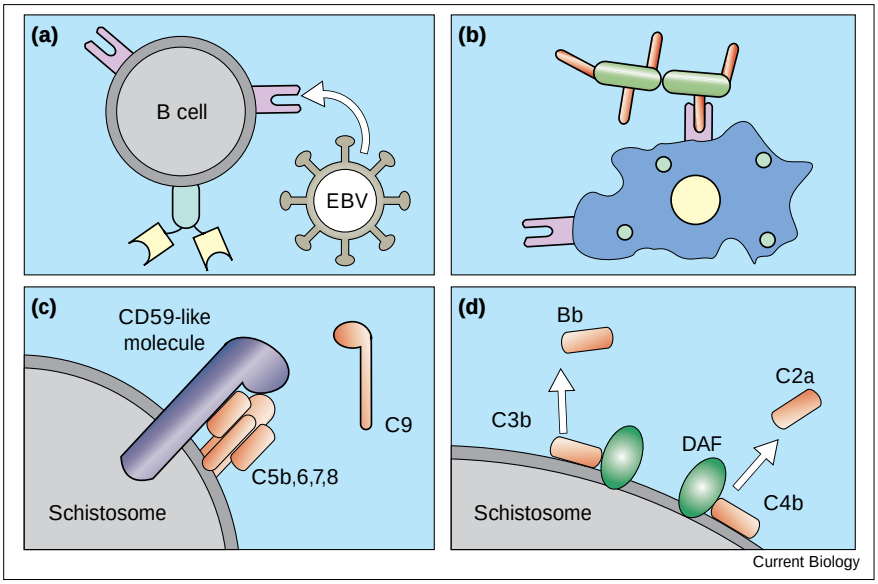
Transplantation of organs between species that are distantly related in phylogenetic terms — such as humans and pigs — is followed by rapid organ rejection, which is mediated by massive complement activation, caused by antibody binding to abundant carbohydrate epitopes on the pig tissues. Because xenotransplantation is an attractive option (if ethical and viral issues permit) for overcoming the severe shortage of human organs for transplantation, there has been much interest in inhibiting complement-mediated injury to xenotransplants. Pigs have been developed that are transgenic for the expression of human complement regulatory proteins. Organs from such animals show significant protection from hyperacute rejection mediated by complement when transplanted into non-human primates.

A clinically-important example of complement-mediated tissue injury is that following infarction. For example, the MAC might be found in the heart following myocardial infarction. Soluble fragments of the complement receptor and regulatory protein CR1 (CD35) reduce the infarct size in experimental models of myocardial infarction. This has yet to be translated into clinical practice, but complement activation in infarction and ischaemic-reperfusion injury remains a very attractive target, especially if a small molecule could be discovered with efficient inhibitory activity for complement.

Conclusion

When one of the authors of this article, MW, entered complement research he was told, possibly tongue in cheek, by a distinguished Professor of Medicine that complement research was dead. We hope this primer will persuade that professor that the complement system is alive and well and that the study of complement continues to make major and surprising contributions to our understanding of immunobiology.

Figure 2



Four strategies by which pathogens use the complement system as part of their pathogenesis. (a) The Epstein–Barr virus (EBV) uses a B lymphocyte receptor for complement, CR2 (pink), to gain entry to the cell. (Cell-surface immunoglobulin is shown in yellow.) (b) A mycobacterium (green) fixes a complement enzyme, C2a, to its surface. The C2a cleaves C3, resulting in the binding of C3b (red) to the mycobacterial surface. The mycobacterium then enters macrophages (blue) via C3 receptors (pink) on the cell surface. (c) Schistosomes synthesize an endogenous protein (purple) that mimics a host complement regulatory protein, CD59, and inhibits the formation of the MAC (red) on the parasite. (d) Schistosomes absorb a complement regulatory protein, decay accelerating factor (DAF; green), from the host plasma that protects against the activation of complement by inhibiting the formation of the C3 convertase enzyme.

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